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Dynamic Model of Heat Inactivation Kinetics for Bacterial Adaptation▽

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The Weibullian-log logistic (WeLL) inactivation model was modified to account for heat adaptation by introducing a logistic adaptation factor, which rendered its "rate parameter" a function of both temperature and heating rate. The resulting model is consistent with the observation that adaptation is primarily noticeable in slow heat processes in which the cells are exposed to sublethal temperatures for a sufficiently long time. Dynamic survival patterns generated with the proposed model were in general agreement with those of *Escherichia coli* and *Listeria monocytogenes* as reported in the literature. Although the modified model's rate equation has a cumbersome appearance, especially for thermal processes having a variable heating rate, it can be solved numerically with commercial mathematical software. The dynamic model has five survival/adaptation parameters whose determination will require a large experimental database. However, with assumed or estimated parameter values, the model can simulate survival patterns of adapting pathogens in cooked foods that can be used in risk assessment and the establishment of safe preparation conditions.

Combined with heat transfer data or models, microbial survival kinetics, especially of bacteria or spores, is extensively used to determine the safety of industrial heat preservation processes like canning, extant or planned. The same is true for milder heat processes such as milk and fruit pasteurization. However, survival models are also a valuable tool to assess the safety of prepared foods, especially those made of raw meats, poultry, and eggs, where surviving pathogens can be a public health issue.

The heat resistance of a bacterium, or any other microorganism, is almost always determined from a set of its isothermal survival curves, recorded at several lethal temperatures. The kinetic models, which define the heat resistance parameters, may vary, but the calculation procedure itself is usually the same. First, the experimental isothermal survival data are fitted with what is known as the "primary model." Once fitted, the temperature dependence of this primary model's coefficients is described by what is known as the "secondary model." When combined with a temperature profile expression, $T(t)$, and incorporated into the inactivation rate equation, the result is a "tertiary model," which enables its user to predict the organism's survival curve under any static or dynamic (i.e., nonisothermal) conditions.

The traditional log-linear ("first-order kinetic") model is the best-known primary survival model, and it is still widely used in sterility calculations in the food, pharmaceutical, and other industries. Traditionally, it has been assumed that the D value calculated with this model has a log-linear temperature dependence or, alternatively, that the temperature effect on the exponential rate constant, k, the D value's reciprocal, follows the Arrhenius equation. However, accumulating experimental evidence in recent years indicates that bacterial heat inactivation only rarely follows the first-order kinetics and that there is no reason that it should (3, 18, 29). Nonlinear survival curves can be described by a variety of mathematical models (6). Perhaps the most frequently used in recent years is the Weibullian model, of which the traditional log-linear model is a special case—see below.

Regardless of the log-linearity issue, none of the abovementioned models accounts for adaptation, the ability of certain bacterial cells to adjust their metabolism in response to stress in order to increase their survivability (2, 10, 26, 27, 28). A notable example is *Escherichia coli*. Its cells can produce "heat shock proteins," which help them to survive mild heat treatments (1, 11). Other organisms, *Salmonella enterica* and *Bacillus cereus* among them, can also develop defensive mechanisms that help them to survive in an acidic environment (8, 9, 13). Whether adaptation allows the cells to avoid injury or to repair damage once it has occurred, or both, should not concern us here. (Injury and recovery, although related, are a separate issue, one which is amply discussed in the literature. Their quantitative aspects and mathematical modeling are discussed elsewhere [5].)

The cells' ability to augment their resistance is not unlimited, and it takes time for the cells to activate the protective system and synthesize its chemical elements (10, 12). Consequently, the effect of heat adaptation on an organism's survival pattern becomes measurable only at or at slightly above what's known as the "sublethal" temperature range. Under dynamic conditions, therefore, adaptation can be detected only when the heating rate is sufficiently low to allow the cells to respond metabolically to the heat stress prior to their destruction.

Several investigators have reported and discussed the quantitative aspects of adaptation (25, 27, 28). When it occurs, adaptation is noticed as a gap between survival

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curves determined at low heating rates and those predicted by kinetic models whose parameters had been determined at high lethal temperatures (7, 8, 9, 27, 28). The question is how to modify the inactivation kinetic model so that it can properly account for adaptation at low heating rates while maintaining its predictive ability at high rates and clearly lethal temperatures. Stasiewicz et al. (25) have recently given a partial answer to this question. They started with the Weibullian inactivation model (see below) and assumed that its rate parameter's temperature dependence follows a modified version of the Arrhenius equation. Using this model and experimental data for *Salmonella* bacteria, they showed that a "pathway-dependent model" is more reliable than a "state-dependent model."

The objectives of our work were to develop a variant of the Weibullian-log logistic (WeLL) inactivation model to account for dynamic adaptation and to demonstrate its applicability with reported adaptive survival patterns exhibited by *Escherichia coli* and *Listeria monocytogenes*, two organisms of food safety concern.

Theoretical background. (i) Weibullian inactivation without adaptation. Published isothermal survival curves of *Escherichia coli*, *Salmonella*, and other pathogens indicate that their heat inactivation does not follow the traditionally assumed first-order kinetics and that their curvilinear semilogarithmic survival curves can be described by the Weibullian model (14, 15, 16, 23, 29).

A convenient way to write this model is.

$$\log_{10} S(t) = -b(T)t^{n(T)} \tag{1}$$

where $S(t)$ is the momentary ("instantaneous") survival ratio, i.e., $N(t)/N_0$, where $N(t)$ and N_0 are the momentary and initial counts and $b(T)$ and $n(T)$ are temperature-dependent coefficients. According to this model, $n(T) > 1$ means that the semilogarithmic survival curve has downward concavity and $n(T) < 1$ means that the curve has upward concavity ("tailing"). The log-linear model is a special case of equation 1 where $n(T) = 1.0$. Thus, the discussion below will be relevant to both linear and nonlinear heat inactivation patterns.

For several organisms, the power $n(T)$ in equation 1 was found to be practically constant or could be assigned a fixed numerical value with only minor effect on the model's fit (6, 14, 16, 22). We will assume that this is true and for what follows use the model with $n(T) = n$, i.e.,

$$\log_{10} S(t) = -b(T)t^{n} \tag{2}$$

The temperature dependence of the "rate parameter," $b(T)$, can be described by the empirical log-logistic model (16, 17, 20):

$$b(T) = \ln\{1 + \exp[k(T - T_c)]\} \tag{3}$$

where T_c marks the temperature level of the inactivation's onset and k is, approximately, the slope of the $b(T)$-versus-T relationship at $T \gg T_c$. According to this model, at $T \gg T_c$, $b(T) \approx k(T - T_c)$, while at $T \ll T_c$, $b(T) \approx 0$, i.e., no measurable inactivation takes place. (Unlike the Arrhenius equation and its variants, the log-logistic model makes a clear distinction between lethal and nonlethal temperatures and it does not require the temperature scale compression [18].)

We assume that under dynamic heating conditions, the momentary inactivation rate is the rate that corresponds to the momentary temperature, at a time that corresponds to the momentary survival ratio (18, 21, 22). Thus, when the inactivation pattern follows equation 2 as a model, the momentary isothermal survival rate, $d \log_{10} S(t)/dt$, at a given temperature T is

$$\frac{d \log_{10} S(t)}{dt} = -b(T)n\, t^{n-1} \tag{4}$$

According to equation 2, the time t^*, which corresponds to the momentary logarithmic survival ratio, $\log_{10} S(t)$, is

$$t^* = \left[-\frac{\log_{10} S(t)}{b(T)}\right]^{\frac{1}{n}} \tag{5}$$

Combining equations 4 and 5 and allowing the temperature to be a function of time, i.e., $T = T(t)$, renders the survival rate equation (18, 21, 22)

$$\frac{d \log_{10} S(t)}{dt} = -b[T(t)]n\left\{-\frac{\log_{10} S(t)}{b[T(t)]}\right\}^{\frac{n-1}{n}} \tag{6}$$

When $b(T)$ is defined by equation 3, equation 6 becomes the WeLL model:

$$\frac{d \log_{10} S(t)}{dt} = -\ln\{1 + \exp\{k[T(t) - T_c]\}\} \cdot n \cdot \left[-\frac{\log_{10} S(t)}{\ln\{1 + \exp\{k[T(t) - T_c]\}\}}\right]^{\frac{n-1}{n}} \tag{7}$$

The WeLL model (equation 7) is an ordinary differential equation that can be solved numerically for almost any conceivable practical temperature profile $T(t)$. We used Mathematica 6 (Wolfram Research, Champaign, IL) for this purpose, but other commercial programs such as Maple or MatLab will also work. (Equation 7 can also be converted into a difference equation and solved with general-purpose software like MS Excel [19].)

The validity of the WeLL model has been demonstrated by its ability to correctly predict dynamic inactivation patterns from experimental isothermal survival data (6, 16, 24) or from dynamic data not used in its parameter calculations (4, 17, 20).

(ii) Adaptation under constant heating rate. Consider an organism whose heat inactivation kinetics follows the WeLL model (equations 2 to 7) being heated under time-temperature conditions that do not allow growth. For simplicity, we will assume that n in the inactivation model is practically unaffected by whether adaptation takes place or not. If this assumption is justified, then adaptation will influence only the magnitude of the Weibullian rate parameter, $b(T)$, making it heating rate dependent.

Let us start with the simple case of constant rate heating, i.e., $dT(t)/dt = v$. In order to express $b(T)$ as a function of the heating rate too, i.e., as $b(T,v)$, we have to take into account the following facts. (i) Adaptation occurs primarily in the sublethal temperature range, i.e., before the onset of the massive mor-

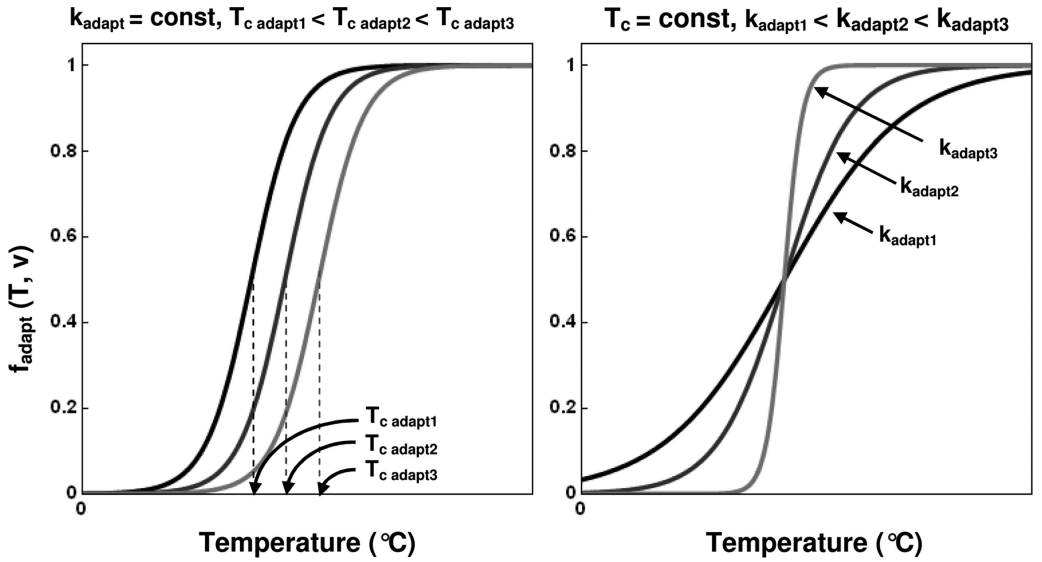

FIG. 1. Schematic view of the construction of the (logistic) adaptation factor, $f_{adapt}(T,v)$, using equation 8 as a model.

tality of the cells. (ii) When adaptation occurs, the lower the heating rate is, the longer the cells remain at temperatures that allow them to activate their protective mechanism(s). Therefore, the lower the heating rate, the greater is the adaptation effect on the organism's survival pattern. (iii) The increased heat resistance caused by adaptation has an upper limit set by physical considerations. In other words, even at rates low enough to be considered isothermal, i.e., where $dT(t)/dt \rightarrow 0$, the organism cannot become absolutely heat resistant. (iv) At high heating rates, the adapted cells' heat resistance approaches that of the unadapted cells. (v) Adapted cells do not become more heat sensitive at high temperature or with fast heating. Note that the above five considerations are not contingent on any specific mechanism(s) operating at the molecular and cellular level. They also do not require that the protective mechanism(s) remain unchanged over the whole pertinent temperature range.

The task now is to find a mathematical expression of $b(T,v)$ whose insertion into the inactivation rate equation will produce survival curves that satisfy the five conditions and for which the resulting model will be consistent with experimental observations. More specifically, the sought-for expression $b(T,v)$, like the original $b(T)$ from which it will be derived, should be zero or approximately zero at low temperatures where no inactivation takes place and sensitive to v only within a certain intermediate (sublethal) temperature range. At high lethal temperatures, where adaptation is impossible regardless of the heating rate, the value of $b(T,v)$ should coincide with that of the original $b(T)$.

A way to formulate such an expression is to multiply the original Weibullian rate parameter $b(T)$ by a logistic adaptation factor, $f_{adapt}(T,v)$, of the kind depicted schematically in Fig. 1., ie.,

$$f_{adapt}(T,v) = \frac{1}{1 + \exp\{k_{adapt}[T_{c\ adapt}(v) - T]\}} \quad (8)$$

In this expression, k_{adapt} is a constant marking the steepness of the adaptation factor $[f_{adapt}(T,v)]$-versus-temperature relationship around the inflection point $T_{c\ adapt}(v)$ (Fig. 1) and which accounts for the increase in heat resistance of the adapted cells expressed in terms of "pushing" the original T_c to a higher temperature, i.e., $T_{c\ adapt}(v) \geq T_c$, as shown in Fig. 2. Note that $T_{c\ adapt}(v)$ must decrease with v. It can be, for example,

$$T_{c\ adapt}(v) = T_{limit} - a \cdot v \quad (9)$$

where a is a proportionality constant and T_{limit} is a marker of the temperature at which the adapted cells start to succumb. Combining the "adaptation factor" (equation 8) with the original "rate parameter" (equation 3) yields its adapted version or the heating rate-dependent Weibullian rate parameter:

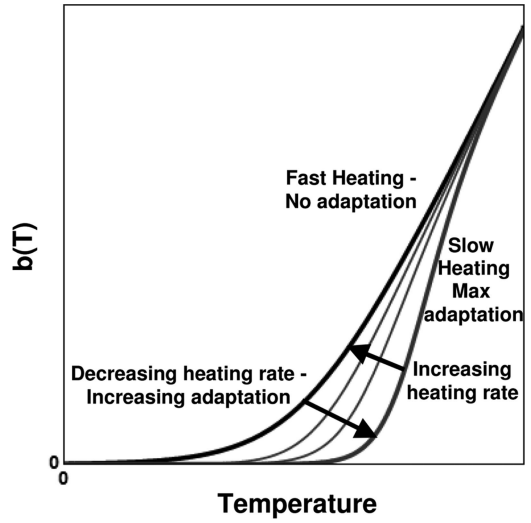

FIG. 2. Schematic view of how an organism's adaptation at sublethal temperatures and low heating rates affects its Weibullian inactivation's rate parameter, $b(T)$. (The curves shown were all produced with equation 10 as a model.)

$$b(T,\nu) = \frac{\ln\{1 + \exp[k(T - T_c)]\}}{1 + \exp\{k_{\text{adapt}}[T_{c\ \text{adapt}}(\nu) - T]\}} \tag{10}$$

Simulated $b(T,\nu)$-versus-temperature and heating rate relationships are also shown in Fig. 2. The figure demonstrates how the expression complies with the stated requirements at least qualitatively. For example, as the heating rate ν increases, the distinction between $b(T,\nu)$ and $b(T)$ progressively disappears. However, even when $\nu \to \infty$, $b(T,\nu)$ cannot be larger than $b(T)$ because the term $1 + \exp\{k_{\text{adapt}}[T_{c\ \text{adapt}}(\nu) - T]\}$ can never be less than 1.0. At the other extreme, even when $\nu \to 0$, $b(T,\nu)$ cannot remain ~0 indefinitely and its value will eventually rise as the temperature increases. According to the simplistic model that we have used for the demonstration (equation 9), it must rise at about T_{limit} or at another definite temperature, had a different $T_{c\ \text{adapt}}(\nu)$ term been used.

When the newly defined rate parameter in the form of $b[T(t),\nu]$ is inserted into equation 7, the result is the inactivation rate model

$$\frac{d\log_{10} S(t)}{dt} = -\frac{\ln\{1 + \exp\{k[T(t) - T_c]\}\}}{1 + \exp\{k_{\text{adapt}}[T_{c\ \text{adapt}}(\nu) - T(t)]\}} \cdot n \cdot \left\{-\frac{\log_{10} S(t)}{\dfrac{\ln\{1 + \exp\{k[T(t) - T_c]\}\}}{1 + \exp\{k_{\text{adapt}}[T_{c\ \text{adapt}}(\nu) - T(t)]\}}}\right\}^{\frac{n-1}{n}} \tag{11}$$

Although equation 11 has an even more cumbersome appearance than equation 7, it is still an ordinary differential equation that can be solved numerically to produce the survival curve $\log_{10} S(t)$-versus-time relationship. Equation 11 can be used to generate nonisothermal survival curves that correspond to a variety of constant heating rates. Examples are given in Fig. 3. They demonstrate how adaptation delays the inactivation's onset at low heating rates and how its effect diminishes as the heating rate increases. Although the simulated survival curves shown in this figure were all created with a model having the simplistic term $T_{\text{limit}} - a \cdot \nu$ (from equation 9) in its formula, they still capture the essence of the adaptation phenomenon and how it shifts the survival curve. Note that a survival curve's shift to the right means increased heat resistance while a shift to the left increases sensitivity. Figure 3 also provides visual demonstration of why an inactivation model derived from survival data obtained at high lethal temperatures fails to predict survival patterns at sublethal temperatures if the organism in question is capable of adaptation. The opposite is also true. An inactivation model for an adaptive organism determined from survival data obtained at low temperatures will fail to predict inactivation patterns at high temperatures and high heating rates. In that case, however, the predicted survival curves will be shifted to the right of the correct ones instead of to the left.

(iii) Adaptation under arbitrary heating rate regimens. If the outlined principles are valid, then they should apply to any monotonic rise in temperature and not only to constant rate heating. However, in order to account for a variable rate, the constant heating rate ν in equation 11 ought to be replaced by the momentary ("instantaneous") heating rate, $dT(t)/dt$, transforming the rate equation into

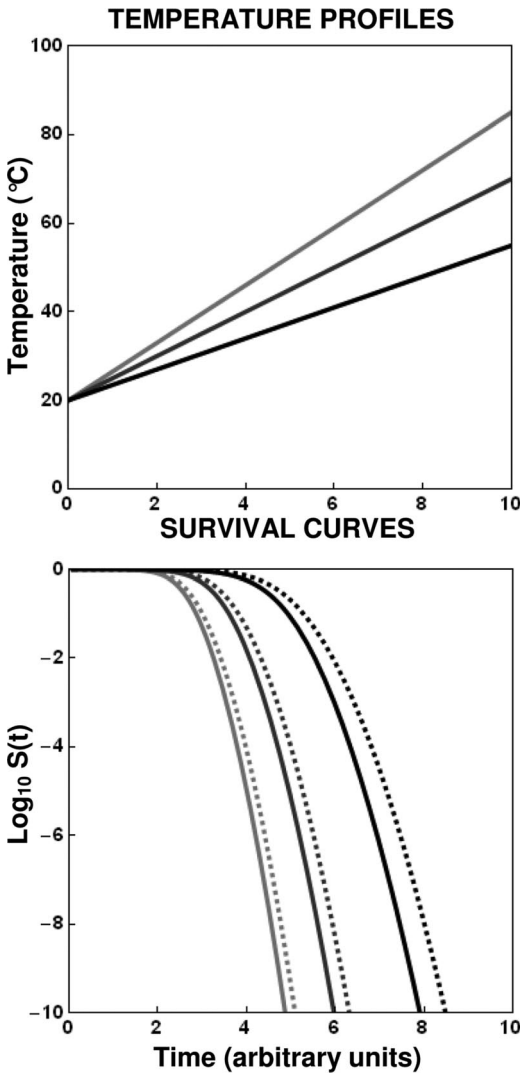

FIG. 3. Simulated constant heating rate temperature profiles and corresponding survival curves. The solid curves were generated with a model that does not take adaptation into account (equation 7), and the dashed lines were generated with a model that does (equation 11). Note that, according to the model, an organism's adaptation is primarily manifested at low heating rates.

$$\frac{d\log_{10} S(t)}{dt} = -\frac{\ln\{1 + \exp\{k[T(t) - T_c]\}\}}{1 + \exp\left\{k_{\text{adapt}}\left[T_{c\ \text{adapt}}\left(\dfrac{dT(t)}{dt}\right) - T(t)\right]\right\}} \cdot n \cdot \left\{-\frac{\log_{10} S(t)}{\dfrac{\ln\{1 + \exp\{k[T(t) - T_c]\}\}}{1 + \exp\left\{k_{\text{adapt}}\left[T_{c\ \text{adapt}}\left(\dfrac{dT(t)}{dt}\right) - T(t)\right]\right\}}}\right\}^{\frac{n-1}{n}} \tag{12}$$

This is a more elaborate model than equation 11, but it too can be solved by Mathematica for a large variety of heating regimens. Examples of temperature histories (profiles) and corresponding survival curves generated with equation 12 as a model are given in Fig. 4. They demonstrate that the rate equation's complexity is no hindrance to its numerical solution by modern mathematical software. They also show that, as the heating rate is increased, the

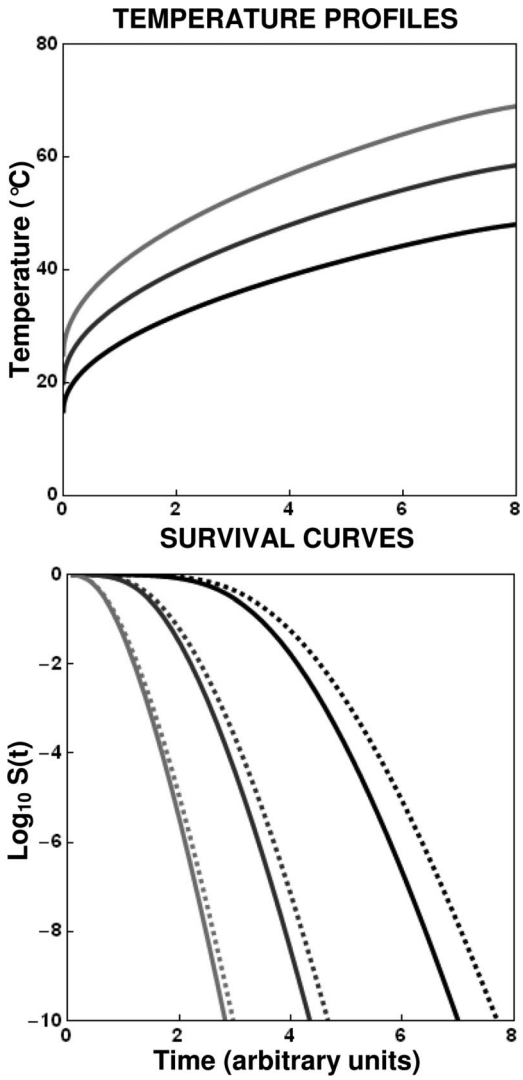

FIG. 4. Simulated variable rate temperature profiles and corresponding survival curves. The solid curves were generated with a model that does not take adaptation into account (equation 7), and the dashed lines were generated with a model that does (equation 12). Note that, according to the model, an organism's adaptation is primarily manifested under slow heating regimens and that in a monotonic temperature rise the inactivation rate equation's complexity does not affect the Mathematica program's ability to solve it numerically.

survival curve of cells capable of adaptation can become indistinguishable from that of cells that are not.

Since the constant rate heating model (equation 11) is just a special case of equation 12 where $dT(t)/dt = v$, the latter can be used for both linear and nonlinear heating. Another special case is an isothermal heat treatment where $dT(t)/dt = 0$, in which case equation 12 is reduced to

$$\frac{d \log_{10} S(t)}{dt} = -\frac{\ln\{1 + \exp[k(T - T_c)]\}}{1 + \exp[k_{\text{adapt}}(T_{\text{limit}} - T)]} \cdot n \cdot \left\{-\frac{\log_{10} S(t)}{\frac{\ln\{1 + \exp[k(T(t) - T_c)]\}}{1 + \exp[k_{\text{adapt}}[T_{\text{limit}} - T]]}}\right\}^{\frac{n-1}{n}} \quad (13)$$

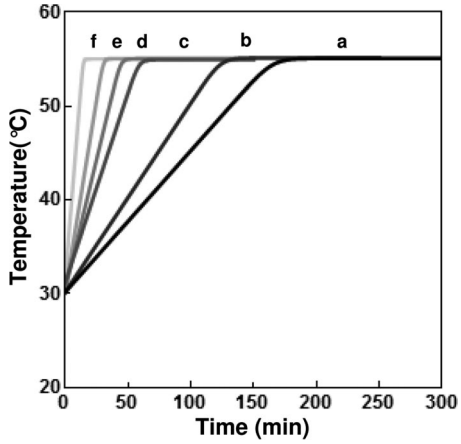

FIG. 5. Six temperature profiles of heat treatments used to inactivate *E. coli*. The original data are from the work of Valdramidis et al. (27).

Comparison of the model with published experimental observations. (i) *Escherichia coli*. Six nonisothermal temperature profiles and corresponding survival data of *E. coli* K-12 MG1655, originally reported by Valdramidis et al. (27, 28), are shown in Fig. 5 and 6, respectively. The treatments consisted of a period of heating at a constant rate to reach 55°C, at which point this temperature was maintained until the process was completed after 6 hours. The difference between the treatments was the heating rate and (consequently) the holding time at 55°C. This kind of temperature profile can be described by the empirical model

$$T(t) = 55 - \ln\{1 + \exp[\nu(t_{55} - t)]\} \quad (14)$$

where ν is the heating rate at the initial stage in °C/min and t_{55} is the time in min to reach the temperature 55°C. The authors of the original publication also determined the organism's isothermal inactivation curves at various temperatures in the range of 52 to 60.6°C, from which its WeLL model's parameters n, k, and T_c could be determined. However, these parameters were determined only from data collected at temperatures higher than 54°C, i.e., lethal to the organism. These were then inserted into the model's equation (equation 7) together with the appropriate temperature profile term (equation 14) in an attempt to predict the survival curve in each of the treatments. The results are shown as solid lines in Fig. 6. Because of the organism's ability to adapt at sublethal temperatures, all the predictions based on the unmodified model were off mark. As expected and as stressed by Valdramidis et al. (28), they were all to the left of the actual survival curve. Also, the magnitude of the shift increased as the heating rate was decreased, again in agreement with the model's prediction. (Note that the time scales of the plots in Fig. 6 are not the same.) Fitting the experimental data by conventional regression methods using equation 12 as a model is not a viable option in this case. This is primarily because the model's equation has five adjustable parameters. The problem is further aggravated by the inevitable experimental scatter. However, by letting n assume a representative value and fixing that of a (equation 9), it was possible to get rough estimates of the other inactivation/adaptation parameters, namely, k, T_{limit}, and k_{adapt}, by using a

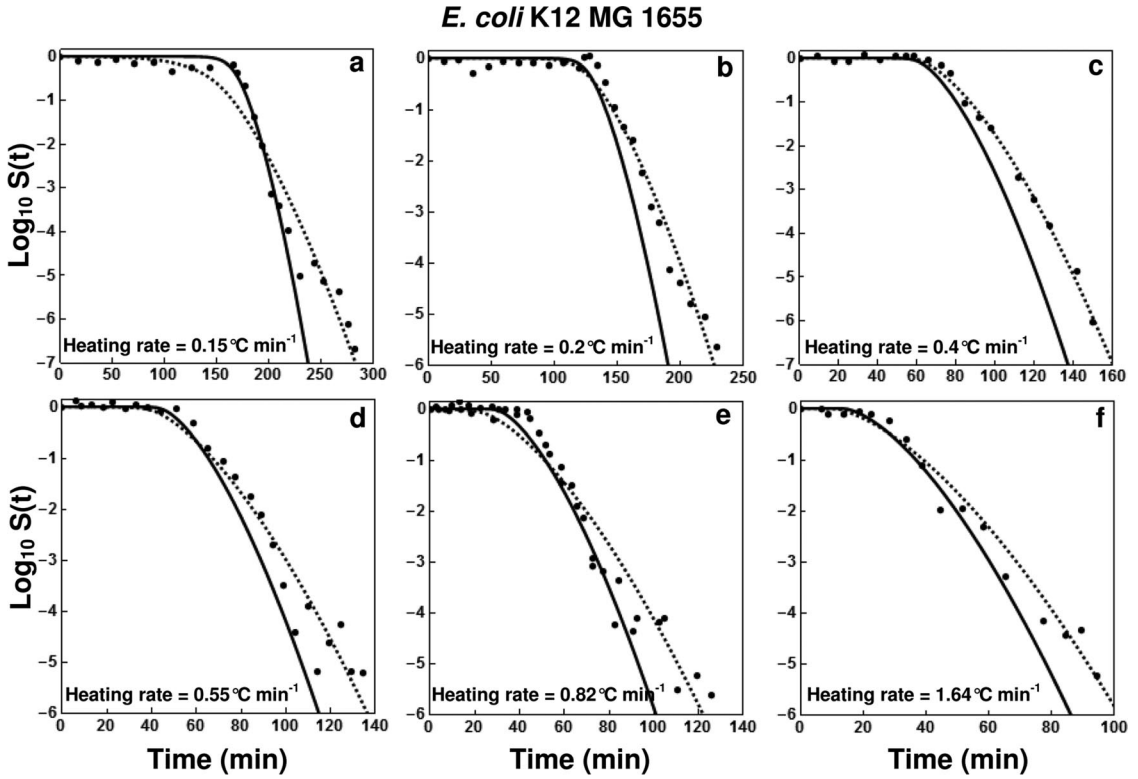

FIG. 6. Survival patterns of *E. coli* K-12 MG1655 exposed to the thermal treatments whose temperature profiles are shown in Fig. 5. The solid curves were generated with a model that does not take adaptation into account (equation 7), and the dashed ones were generated with a model that does (equation 12). The experimental data shown are from the work of Valdramidis et al. (28). Note that the time scales of the survival curves are different.

minimization method offered by Mathematica. They were inserted into the model equation (equation 12) to produce the survival curves corrected for adaptation, which are shown as dashed lines in the figure. (The parameters' approximate values were $a = 2.0$ to 2.5 min, $n = 1.6$, $k = 0.02$ to $0.3°C^{-1}$, $T_{limit} = 58$ to 84°C, and $k_{adapt} = 0.2$ to $1.3°C^{-1}$ with two clear outliers.) Because alteration of one parameter in order to minimize the mean square error is compensated for by changes in the other parameters' magnitudes, some of the estimates varied within a wide range. However, despite these limitations and the grossly oversimplified assumption that the adaptation could be accounted for by equation 9, the "correct survival curves" were at least visually in agreement with those actually observed (Fig. 6). In other words, despite the crudeness of the model and its parameter estimation procedure, it still captured the essence of the organism's adaptation and how it is manifested in slow and fast heating regimens.

(ii) *Listeria monocytogenes*. Three constant heating rate temperature profiles and corresponding survival data for *L. monocytogenes* at pH 7.4 and 5.5 are shown in Fig. 7 and 8, respectively. The original authors of the data are Hassani et al. (8, 9, 10), who also reported the organism's isothermal survival patterns at these two pH levels (10). As before, the WeLL model's parameters were calculated using data obtained at clearly lethal temperatures, i.e., in the range of 54 to 62°C. These were inserted into equation 7 with the temperature profile terms to produce the survival curves shown as solid lines in Fig. 8. As in the case of *E. coli*, all the curves predicted without taking adaptation into account were to the left of the ones observed experimentally. Also, as expected, the gap between them decreased as the heating rate increased. (Again, note that the time scales of the plots are very different.) This was observed at both pH levels. As could be expected, lowering the pH intensified the heat's inhibitory effect, but the data at hand were

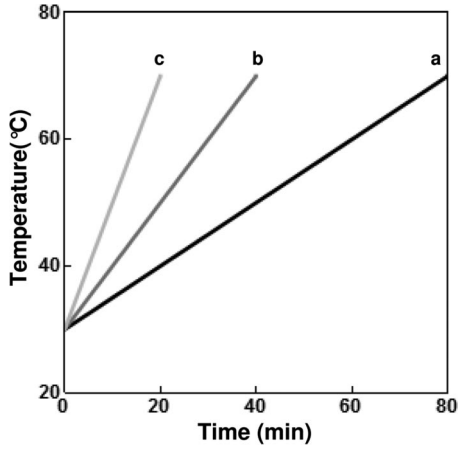

FIG. 7. Three temperature profiles of heat treatments used to inactivate *Listeria monocytogenes.* The original data are from the work of Hassani et al. (8).

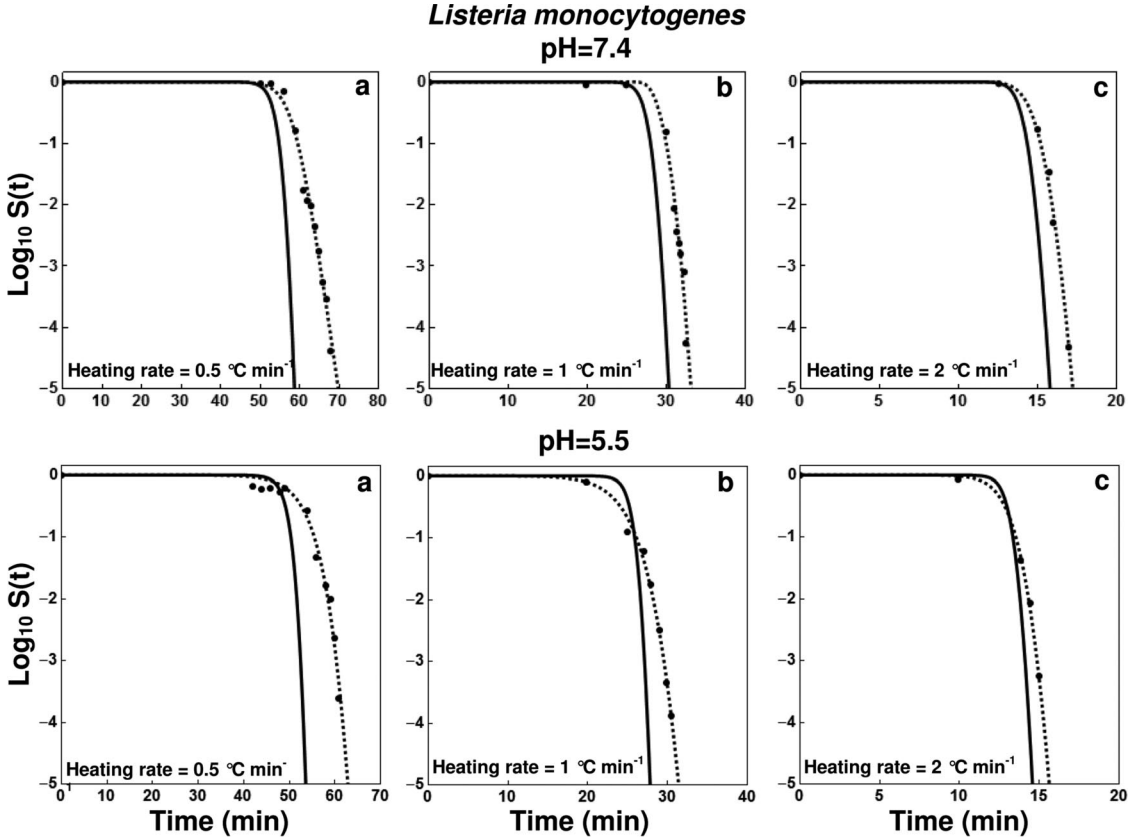

FIG. 8. Survival patterns of *Listeria monocytogenes* exposed to the thermal treatments whose profiles are shown in Fig. 7. The solid curves were generated with a model that does not take adaptation into account (equation 7), and the dashed ones were generated with a model that does (equation 12). The original data are from the work of Hassani et al. (8). Note that the time scales of the survival curves are not the same.

insufficient to determine whether it also had a significant effect on the organism's adaptation pattern.

As before, setting n at a representative value and fixing that of a enabled us to estimate the other model parameters with a minimization procedure. Once they were estimated, we could generate the survival curves corrected for adaptation using equation 11 as a model. The approximate values of the parameters were $a = 2\ \text{min}^{-1}$, $n = 0.8$ for pH 7.4 and 1.1 for pH 5.5, $k = 0.3$ to $0.7°\text{C}^{-1}$ with one clear outlier, $T_{\text{limit}} = 56$ to 60°C, and $k_{\text{adapt}} = 0.1$ to $0.8°\text{C}^{-1}$ with one clear outlier. The survival curves corrected for adaptation are shown as dashed lines in Fig. 8. In all six cases, they were in almost perfect agreement with the actual data. The calculated values of k and k_{adapt} varied considerably and showed no trends. T_{limit}, in contrast, varied within the narrow range of 56 to 60°C, which is probably consistent with the upper limit of the organism's sublethal temperature range.

Perhaps with the exception of T_{limit}, the exact meaning of the estimated parameters' magnitudes is unclear at this point. However, they might indicate their expected order of magnitude. Like those of *E. coli*, the published experimental *Listeria* data used in the analysis had not been originally intended to test the proposed adaptation model. Yet, the same model that fitted the *E. coli* data also fitted the six experimental survival curves of *Listeria*. This suggests that the proposed mathematical model indeed captured the two organisms' adaptation patterns despite the different concavities of their isothermal survival curves, i.e., $n = 1.6$ versus 0.8 or 1.1, respectively. It should be added that conventional statistical fit measures were not applicable here and are therefore not reported. This is because one of the parameters, a, was adjusted. Also, the deviation of the corrected curves from those where adaptation had not been taken into account was systematic rather than random and its magnitude varied with the rate according to the prediction of the model itself.

Concluding remarks. Pathogen adaptation has a direct impact on the safety of foods, especially if they are only marginally cooked at home or by vendors. Consequently, quantifying the adaptation effect on the survival curve is of prime importance to public health. In this work, we have demonstrated that the WeLL model can be modified to account for heat adaptation and that dynamic survival curves produced by this modified version are qualitatively consistent with those of *E. coli* and *L. monocytogenes* as reported in the literature. With adjustment of its parameters, the modified model could also be made to agree quantitatively with the experimental survival data. This suggests that it is possible, at least in principle, to quantify the role of adaptation in terms of the parameters of an inactivation rate model. In the described model, these were T_{limit}, k_{adapt}, and a defined by equations 8 to 10, but similar parameters could be derived for alternative models. The inactivation/adaptation model that we have chosen and used is

perhaps the simplest possible one, since we have assumed that T_{limit} decreases linearly with the heating rate. Yet, even with this simplistic assumption, the inactivation rate model (equation 11 or 12) had five survival/adaptation parameters, namely, a, n, k, T_{limit}, and k_{adapt}. Reliable determination of all five would require the creation of a large experimental database, which currently does not exist. For this reason, the described model could be evaluated only by its qualitative predictions. However, by its parameter adjustment, the model could be used to fit experimental survival curves in which adaptation played a variable role, depending on the heating rate and temperature range. This suggests that the described model with assumed parameters can be used to simulate survival curves of adapting pathogens under conditions that emulate realistic scenarios such as those that exist in cooking or grilling. Such curves could then be used in risk assessment of current practices, could assist health authorities to determine safe cooking conditions, and could be used to assess the potential safety implications of emerging adaptation in existing pathogens or the appearance of new ones. As far as computation is concerned, the complexity of the inactivation/adaptation rate model (equation 11 or 12) is no hindrance to its application. The differential equation can be rapidly solved with software like Mathematica to produce the survival curve under practically any monotonic heating regimen. The same would probably be true if a more elaborate model were employed to account for the adaptation phenomenon.

This communication focuses on a situation where the pathogen's population, real or contemplated, is subjected to heat as the means of its destruction, in which case the survival curve would indicate how effective the treatment is. The methodology, however, can be extended to other means of bacterial inactivation or suppression, such as chemical disinfection and refrigeration or freezing. An important issue not addressed in this paper is the fate of adapted and unadapted cells if and when the heating is terminated prematurely, allowing the survivors to grow. This too might be an issue of food safety concern, but its investigation and modeling require a different kind of experimental data and mathematical models. To account for the inactivation/adaptation kinetics, the discussion focused on what happens at a point. However, once the kinetics has been established, it could be combined with heat transfer theories in order to calculate the number of surviving pathogens in a volume of food undergoing insufficient or marginal heat treatment.

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